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RESEARCH AND UTILIZATION OF AMORPHOPHALLUS IN CHINA *

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Abstract In this article, the authors reviewed systematically the research works on konjac conducted mainly by the Research Center of Konjac, Southwest Agricultural University as well as other organizations in the past 15 years including the resources, biology, agronomy, biochemistry, medical science and usage of konjac. There have been about two thousand years of cultivation of konjac (elephant - foot yam) in China, but such cultivation was fragmentary and scattered around the growers' house, and the use was merely limited to konjac curd for food. It was not until the 1980s that the production of konjac on a large area of land increased up quickly. The acreage under *Amorphophallus* (konjac) reached about 30,000 hectares. A new industry on a large scale was formed following the progress in agriculture and processing. The research work advanced vigorously. Scientific technology is promoting the development of the industry.

Key words *Amorphophallus*, China, research, utilization

GERMPLASM RESEARCH OF *AMORPHOPHALLUS*

1. Species and Their Distribution of *Amorphophallus* in China

There are about 170 species of *Amorphophallus* Blume ex Decaisne in the world (Mayo *et al*., 1997). Twenty one of them were recorded in China, 12 species have been discovered and described or recorded since 1980: *A. yuloensis* H. Li, *A. pingbianensis* H. Li et C. L. Long, *A. nanus* H. Li et C. L. Long (named by Li Heng *et al*. at the Kunming Institute of Botany); *A. hayi* W. Hett., *A. arnautovii* W. Hett., *A. odoratus* W. Hett. et H. Li (named by Wilbert Hetterscheid from the Netherlands); *A. albus* Liu et Chen (named by Liu Peiying *et al*. at Southwest Agricultural University); and *A. coaetaneus* Liu et Wei (named by S. Y. Liu *et al*. at Guangxi Traditional Chinese Medical College), while *A. krausei* Engl., *A. corrugatus* N. E. Brown, *A. kachinensis* Engl. et Gehrm. and *A. tonkinensis* Engl. et Gehrm. are new rocords in China. The distribution of *Amorphophallus* in China extends southwest from Qinling (a mountain range in south Shanxi). There is a gradual decrease of germplasm resources with the increase of latitude (Li, 1988; Yang *et al*., 1990) (Table 1). Ten of them are endemic to China. They are *A. hirtus* N. E. Brown, *A. henryi* N. E. Brown, *A. dunnii* engler, *A. stipitatus* Engler, *A. mellii* Engler, *A. coaetaneus* S. Y. Liu et S. J. Wei, *A. albus* P. Y. Liu et J. F. Chen, *A. yuloensis* H. Li, *A. nanus* H. Li et C. L. Long and *A. odoratus* W. Hett. et H. Li (Li and Long, 1998).

2. Germplasm Research of *Amorphophallus* in China

(1) Studies on Palynology

Gong *et al*. (1990) studied 10 species of *Amorphophallus* and noted that the pollen is large and has thick exines, which are particular to the Araceae. Based on the sculpture of pollen exines, two groups were divided. One is cerebriform, the other striped form. The pollens of *A. konjac* K. Koch and *A. nanus* belong to the former while *A. yunnanensis*, *A. dunnii*, *A. albus*, and *A. krausei* are under the latter. In the striped form group, however, the spadix is smaller and a little introversive and encloses the inflorescence. Comparing the thickness of the exine and the ratio of long to short diameter of pollens, *A. dunnii* is much

* Chen Jingfeng, Sun Yuanming, Su Chenggang, Huang Danfeng, Yang Daiming, Wang Yulan, Gong Xianyou, Yang Zhongcheng, Feng Xuqiao *et al*. joined this work

closer to *A. yunnanensis*. However, *A. albus* is much closer to *A. krausei*. *Amorphophallus dunnii* and *A. yunnanensis* are also much alike in plant morphology and different only in the relative length of inflorescence to spadix. Moreover, *A. albus* and *A. krausei* also closely resemble each other and both have sterile neutral flowers in the inflorescence. However, pollen morphology appears to be relatively stable in plant evolution. Whether such differences of pollen sculpturing can be considered as a classification criteria for species in *Amorphophallus* deserves further investigation.

Table 1. Germplasm of *Amorphophallus* in Geographical Demarcation of China

Geographical Demarcations		No. of Species	Species
N. Latitude 35° ↑	Qinling Range Area	1	*konjac*
	Mountain area surrounding Sichuan Basin	3	*konjac*, *albus*, *dunnii*
	Hilly areas of Yangtze and Huai River Valley	2	*konjac*, *kiusianus*
	Hilly areas south of Yangtze River	4	*konjac*, *kiusianus*, *albus*, *dunnii*
	Plateau of Yunnan and Guizhou	8	*kiusianus*, *dunnii*, *konjac*, *yuloensis*, *yunnanensis*, *paeoniifolius*, *albus*, *krausei*
	Hilly areas in Guangdong and Guangxi	10	*konjac*, *yunnanensis*, *paeoniifolius*, *dunnii*, *mellii*, *stipitatus*, *kiusianus*, *yuloensis*, *coaetaneus*, *corrugatus*
	Mountain area of Taiwan	4	*hirtus*, *henryi*, *konjac*, *paeoniifolius*
20° ↓	Paratropical zone of SW Yunnan	7	*konjac*, *yunnanensis*, *paeoniifolius*, *yuloensis*, *pingbianensis*, *yuloensis*, *nanus*, *krausei*

Table 2. Pollen Features in *Amorphophallus*

Source: Gong, 1990

Group	Species	Size (μm)	Ratio of long to short diameter	Shape*	Trait of the Exines			
					Layers	Thick - ness (μm)	Veins	
							photo microscopy	electron microscopy
Cerebri form	*konjac*	43.0 × 39.3	1.10	SS	2	5.4	crevice	cerebriform
	nanus	45.8 × 41.2	1.12	SS	2	3.3 - 5.4	crevice	cerebriform
Striped form	*dunnii*	49.8 × 35.2	1.44	SP	2	4.4	fine stripe	smooth stripe
	yunnan - ensis	55.4 × 37.4	1.49	SP	2	3.3	fine stripe	smooth stripe
	albus	53.0 × 40.9	1.30	SP	2	3.3 - 4.4	fine stripe	smooth stripe
	krausei	55.3 × 44.5	1.25	SP	2	3.3 - 4.4	fine stripe	smooth stripe

* SS = subsphaeroidal; SP = subprolate.

(2) Studies on the Number of Chromosomes and Karyotypes

The number of chromosomes and karyotypes have been studied in several Chinese species of

Amorphophallus since 1985 (Table 3).

(3) Studies on the Chromosome Band Patterns

Zheng *et al*. (1989) studies Giemsa C – band patterns in 4 species of *Amorphophallus*. The results show that C – band is the basic band, which appears consistantly on both long and short arms. W – band and T – band appear less frequently especially the latter. Those bands make up four patterns, i.e. C/C, W/C, C/W and TC/C. (Table 4).

Table 3. Karyotypes of the Main Species in Chinese *Amorphophallus*

Species	Karyotype	Origin	Reference
konjac	2n = 2x = 26 = 16m + 2sm + 8S + (2ST)	Sichuan	Liu Peiying *et al*., 1985
	2n = 2x = 26 = 18m + 16sm + 2st	Yunnan	Long Chun – lin *et al*., 1989
	2n = 2x = 26 = 16m + 10sm	Hunan	Long Chun – lin *et al*., 1989
	2n = 2x = 26 = 6m + 10sm + 10st	Hunan	Zheng Suqiu *et al*., 1989
	2n = 2x = 26 = 16m + 10sm(/st)(2ST)	Sichuan	Gong Xianyou, 1990
albus	2n = 2x = 26 = 16m + 6sm(2ST) + 4st	Pingshan, Sichuan	Liu Peiying *et al*., 1985
	2n = 2x = 26 = 14m + 8sm + 4st	Kunming, Yunnan	Zheng Suqiu *et al*., 1989
	2n = 2x = 26 = 20m(2ST) + 6sm	Jinyang, Sichuan	Li Heng *et al*., 1990
	2n = 2x = 26 = 16m + 8sm + 2st	Pingshan, Sichuan	Gong Xianyou, 1990
dunnii	2n = 2x = 26 = 4m + 14sm + 8st	Hunan	Zheng Suqiu *et al*., 1989
yunnanensis	2n = 2x = 26 = 26m	Yuanjiang, Yunnan	Li Heng *et al*., 1990
nanus	2n = 2x = 26 = 20m + 4sm + 2st	Gejiu, Yunnan	Long Chun – lin *et al*., 1989
krausei	2n = 2x = 26 = 20m + 4sm + 2st	Ximeng, Yunnan	Li Heng *et al*., 1990
	2n = 2x = 26 = 22m + 4sm	Ximeng, Yunnan	Gong Xianyou, 1990
pingbianensis	2n = 2x = 26 = 20m(2ST) + 6sm	Pingbian, Yunnan	Long Chun – lin *et al*., 1989
yuloensis	2n = 2x = 26 = 22m + 4sm	Yunnan	Li Heng *et al*., 1990
kachinensis	2n = 2x = 26 = 22m + 4sm	Banna, Yunnan	Li Heng *et al*., 1990
paeoniifolius	2n = 2x = 28 = 2M + 16m + 8sm + 2st	Banna, Yunnan	Long Chun – lin *et al*., 1989

Gong (1990) studied the chromosomes banding in *Amorphophallus*. Actively – growing root tips of 0.5 cm were pretreated in 1:1 solution of 0.05% colchicine and 0.02M 8 – hydroxyquinoline at 25℃ in the dark for 3 hours. The chromosome samples were made with F – SG method, dried in air for 1 week, and induced C – band, N – band and G – band with different means. The time stained in 3% Giemsa is 15 minutes.

Table 4. Formula of C – band patterns in 4 species of *Amorphophallus*

Species	Formula
A. *konjac*	2n = 2x = 26 = 14C/C + 6W/C + 6C/W
A. *dunnii*	2n = 2x = 26 = 16C/C + 8W/C + 2C/W
A. *albus*	2n = 2x = 26 = 10C/C + 12W/C + 2C/W + 2TC/C

a. C – band

BSG and HSG methods failed to induce C – band on the chromosomes of *Amorphophallus*, but it was successful when the chromosomes were treated in 0.1N NaOH at room temperature or 10 min for at 60℃ for 4 – 5min., and stained in Giemsa after washing in water.

b. N – band

N – band was induced when treated in 1 M NaH_2PO_4 at 96℃ for 40 min. If the treating time was too short, no band was observed, and the band disappeared otherwise.

c. G – band

The samples of chromosomes from *A. konjac*, dried in the air for several days and stained in Giemsa directly, displayed abundant bands of 4 to 11 on every chromosome of late prophase and early metaphase.

When the chromosomes were treated in 6M urea for 5 min; rich and clear G – bands were also acquired.

(4) Studies on Soluble Protein

The soluble protein extracted from seeds of *A. konjac* or *A. albus* was analyzed through PAGE electrophoresis. The results showed that there are 12 bands from the seed storage protein of *A. albus*. The RF value of PRf9 equals 0.2216 and that of PRf12 in 0.9286. In *A. konjac* 9 bands are much similar to those from *A. albus* but the PRF9 is very sensitive to temperature. According to the theory of P. M. Smith, the storage protein in seed, fruit and other organs can served as an index of classification, and difference between species is due to accumulation of mutation. Therefore, it is possible that the divergence in storage protein banding patterns between *A. albus* and *A. konjac* resulted from gene mutations, and the similarity in 9 bands can be regarded as evidence of a close relationship between these two species (Liu *et al*., 1985).

The germplasm research presented above confirmed that different species hold different karyotype. However, for the same species results were inconsistent because of different materials and authors. Presumably such diversity in karyotypes in the same species is caused by environmental adaptation in different regions during a long time or experimental error and standards of different authors.

Karyotype and chromosomes banding patterns can be used to distinguish species, but it is rather difficult to expound the consanguinity relationship and degree of evolution. The main reason is because the experiment results are not very consistant. According to our investigation in cultivated or wild species collected from various regions of China, we believe that to understand the relationships and systematic positions. It is necessary to analyze the many – sided indices synthetically. For example, it is believed that the two most important species in China, *A. konjac* and *A. albus*, have a close relationship, based on morphological observation in many regions and years and on investigations of pollen characteristics, unsymmetrical coefficient of karyotype, and electrophoretic banding patterns of seed storage protein. *Amorphophallus albus* is more primitive. Their affinity was confirmed by reciprocal crosses and fertile F1 hybrid seed were obtained. We also observed that there were consistant relationships between karyotype, pollen characteristics, and morphology. For instance, the pollen of *A. albus*, *A. krausei*, *A. dunnii*, and *A. yunnanensis* are belong to striped type. Their chromosomes have low unsymmetrical coefficients. Those species are believed to be more primitive. Plants are small and have weak growth potential and the spadix is not differentiated. Of these, *A. yunnanensis* is related more closely to *A. dunnii*. *Amorphophallus albus* is closely related to *A. krausei* which both possess a segment of neutral sterile flowers in the inflorescence. On the other hand, the pollen of *A. konjac* and *A. nanus* are cerebriform, the unsymmetrical coefficients of chromosomes are larger, and the spadix is more differentiated. They belong to more advanced species, and have a strong growth potential.

RESEARCH ON BIOLOGY AND BIOCHEMISTRY OF *AMORPHOPHALLUS* PLANTS

1. Growth and Development of Konjac Plants

(1) The Trend of Plant Growth

According to experiments conducted by Chen *et al*. (1983) in Sichuan, *A. albus* pullulated and emerged seedlings on May 20. The leaf developed rapidly, the corm and rhizomes began to grow, and at the same time the seed – corm decreased in weight rapidly.

The corm change stage occurred in early July. In this stage, the seed corm withered and detached from the plant thus ending the seedling stage. The plant hereafter grew vigorously and independently, and the new corm expanded rapidly until late August. This is called the corm rapid – expanding stage. In this stage, the chlorophyll content rose from 0.5mg/g in seedling stage to 1.5mg/g and catalase rose from 2100 $\mu g/min \cdot g^{-1}$ to more than 3200 $\mu g/min \cdot g^{-1}$. Then the growth of leaf, corm and rhizomes slowed down, and the corm weight did not increase anymore until the end of September, at which time the corm was mature.

(2) Flower Bud and Leaf Bud of Corm and Corm Dormancy

Generally, the flower bud differentiates from the terminal bud of four year old *A. konjac* or three year old *A. albus*. Because of the extremely strong apical dominance, once the terminal bud blossoms, none of the axillary buds (leaf buds) pullulates, the "flower and leaf of a same plant never meet". However, if the flower bud is removed early in the growth stage, one of the axillary buds can develop into a leaf in the same year.

Sun (1995) discovered that when the main bud pullutates, endogenous bud differentiates on the stem at the bottom of the leaf stalk and becomes the main terminal bud of the next year. This new bud may remain to be a leaf bud and is dormant from July to the next spring. However, it can also differentiate into a flower bud from about mid – August to late October if the plant is more than 3 or 4 years old. The flower bud does not become active and blossom until the next March to April (*A. konjac*) or May to June (*A. albus*).

The dormancy of corms of konjac is a physiological one. No factor can break corm dormancy and lead to pullulation in its dormant stage.

2. Research on Embryology

The seed of konjac within the fruit is not the true botanical seed but is a kind of corm, according to an investigation of the embryology of *A. konjac* conducted by Zhao *et al*. (1987). The development of the male and female gametophytes is normal. An eight nucleus embryo sac has been clearly seen. The interval between pollination and fertilization is about 2 days and double fertilization proceeds normally. The zygote begins to divide 5 – 6 days later. The first division is transverse to form a two cell proembryo, but the following divisions are not in sequence. Finally, a polycell embryo is formed, and the cell near the micropyle becomes active merismmatic tissue, leading to the formation of a kind of scade – bud. So the embryo of *A. konjac* is single – polar (micropyle), and the activity of the other polar (chalaza) disappears. With the destruction of the endosperm, the interior of the integument is filled by the procorm. The peripheral cells of the procorm carry out periclinal divisions to form the structure from which the cork originates. Almost one month later the little corm is formed. In the entire period of the development of procorm from zygote no structure of gemmule, primordial root and mesocotyl have been seen. The above facts show that the little corm within the fruit is still derived from sexual reproduction.

3. Ecological Conditions

(1) Light

Chen *et al*. (1984) determined that light saturation point of *A. albus* was 17 klx, 22 klx and 18 klx on July 2, August 2 and September 2 respectively. Plants grew most vigorously in August and adapted to a relatively large light intensity range. Photosynthesis efficiency apparently does not drop even in 35 klx light intensity. All light compensation points of konjac are 2 klx and do not change much.

Table 5. Comparison of Photosynthetic Capacity

Species	Temperature (℃)	Net Photosynthetic Intensity ($mgCO_2/dm^2 \cdot hr$)	Respiration Intensity ($mgCO_2/kg \cdot hr$)
A. albus	31	6.45	1.75
A. konjac	31	12.92	1.71
Colocasia esculenta	31	16.56	3.06

It is obvious that photosynthetic efficiency of konjac is relatively low (Table 5). Net photosynthetic intensity of *A. albus* is 6.45, that of *A. konjac*, 12.92. These values are one – half and one – third respectively lower than *Colocasia esculenta* of the same family.

Light intensity affects growth and disease – resistance of konjac. When light intensity rises from 2254 lx to 46000 lx, plant height, growth of leaf and chlorophyll content drop accordingly but disease – destroy rate increases (Table 6). High output is available by proper shading.

(2) Temperature

Zhang *et al*. (1991), found that konjac prefers warm weather. The optimum temperature range for plant growth is 20 – 25℃, and 5 – 43 ℃ is the adaptable temperature range. When exposed to below 0℃ or above 45℃, plants die after 5 days. Bud pullulates and grows when it is above 15℃ in spring, and the leaf withers and lodges when it is below 15℃ or above 35℃ , and the green color of the leaf fads. The permeability of the cell membrane is 62.98%, 3 times higher than that of CK when It is above 45℃. The temperature range of 22 – 26℃ is best for konjac root development, and 22 – 30℃ is best for corm expansion. The corm can be kept in the best condition when the storage temperature is 10℃. When It is below 0℃, cell structure is destroyed and the corm do not pullulate any more. Konjac growth period is affected by cumulative temperature. When the cumulative temperature is high enough, the leaf lodges. Different species require different cumulative temperature. When grown in lower mountainous areas below 800 m in Jinshajiang valley, *A. albus* needs active cumulative temperature 4831℃ and efficient cumulative temperature of 1658℃ from pullulation to lodging. *Amorphophallus konjac* needs active cumulative temperature of 4279℃, efficient cumulative temperature of 1089℃, and grows best in mountainous areas from 800 m to 2500 m.

Table 6. Effects of Shading on the Growth of Konjac

Layer of shading material	Light intensity (lx)	Soil surface temp. (℃)	Plant height (cm)	Width of leaf stretch (cm)	Chlorophyll (mg/gfw)	Disease – destroy rate (%)	Coefficient of yield increase
3	2254	36	38	75	2.02	12.50	0.41
2	4002	35	33	62	2.40	18.75	0.53
1	14674	34	30	50	1.62	25.00	0.16
CK	46000	45	16	34	1.30	87.50	– 0.64

(3) Moisture

Konjac does not require much water, but due to its shallow root system, it has poor drought tolerance. Zhang *et al*. (1991) found that during its vigorous growing early and middle phase, 75% of the field water capacity favors root development. When the corm matures, water content should drop to 60% of the field water capacity. Konjac grows poorly in stagnant. When soil moisture reaches saturated water content, aeration is reduced and the roots die. But if the moisture drops to 25% of the saturated content, the roots and root hairs die completely. The water deficit causes a progressive decline in chlorophyll and protein content in the leaves.

4. Biochemistry of konjac

(1) Konjac glucomannan (KGM) content

Amorphophallus contains mainly starch and KGM, their concentrations are negatively correlated (Table 7). KGM is more valuable. Usually KGM content is 0 to 70%. KGM exists in the corms and some can be found in the leaves. KGM is found in the idioblasts, with the diameter of 0.5 to 2 mm, 10 to 20 times larger than that of starch granule. Each idioblast contains one granule of KGM and is embedded by parenchyma cells with starch granules.

(2) Substance Trends in Growth Period of Konjac Plant

From Table 8, the trends of the main components of corms in different growth periods are obvious:

Table 7. The Components by % of the dry wt of Several Chinese *Amorphophallus* (Sun *et al.*, 1988)

Species	Total Sugar	KGM	Starch	Soluble Sugar	KGM: Starch
A. konjac (Wanyuan)	76.0	59.6	12.5	2.9	4.76:1
A. konjac (Qijiang)	74.8	59.4	12.5	2.9	4.75:1
A. konjac (Pinglan)	77.2	52.7	20.1	3.4	2.62:1
A. albus	72.2	59.2	11.4	1.5	5.20:1
A. kiusianus	71.7	56.7	12.4	2.6	4.573:1
A. krausei	64.4	27.4	34.3	2.7	0.790:1
A. nanus	63.4	29.2	32.2	2.0	0.901:1

a. Seed – new corm exchanging period soluble sugar and protein contents of seed corm increase rapidly with bud pullulation and decline rapidly after 50 days. The KGM and starch of the seed corm are exhausted. Seed corm withered and detached from the new corm two months later. The new corm has already formed in the meantime, but it grows slowly, with fresh weight 4.5% and dry weight 5% of that of the whole growing period.

b. New corm rapid expansion stage. From early July to early August, approximately 30 days, the corm becomes five times larger. Dry material, KGM and starch percentage increase rapidly and reach 20.2%, 51.8%, 10.7% respectively, nearly reach their stable content.

c. New corm continuing expansion and repletion stage from early August to the end of September, leaf growth maximizes. Most photosynthetic products are transported and accumulate in the corm. Fresh weight and dry material occupy 63% and 53% of that of the whole growing period of the new corm. This stage is the critical and determines the yield and quality of corms.

d. New corm mature stage. From the end of September to the end of October, the above ground part gradually withers and turns down, meanwhile the corm becomes mature. The increase of dry weight declines. Other contents change stop changing.

(3) Physiological and Biochemical Changes during Corm Storage

Liu *et al.* (1990) divide the storage stage of konjac corm into four stages. The four stages and their respective physiological and biochemical changes are as follows:

a. Post – harvest ripening stage, from late October to late November. Respiration coefficient reaches 13.7 $mgCO_2 \cdot kg^{-1} \cdot hr^{-1}$. The activity of polyphenol oxidase, catalase and amylase are strong.

b. Dormant stage, from late November to early January. Respiration weakens, activity of various enzymes

are weak, corm enters deep dormancy.

c. Bud pullulation stage, from early January to late February, about 2 months. Respiration remains weak. No detectable change of mannan and amylase activity occurs, but activity of catalase decreases and activity of polyphenol oxidase increases slightly. If it is above 15℃, corm pullulates but does not grow and remains in a relatively quiescent stage before dormancy break.

d. Bud elongation stage, after late February. The activity of polyphenol oxidase and amylase increase, if temperature permits corm "germination" and root growth. Table 9 compares the ingredients changes of the unstored corms and the stored corms.

Table 8. *A. konjac* Yield Formation of Single – plant and Trends of the Changes of Its Main Components (Wang, 1989)

Determining Date (date/mon.)		10/5	30/5	10/6	30/6	10/7	30/7	10/8	30/8	10/9	30/9	10/10	30/10
Developing Time (days)		0	20	30	50	60	80	90	110	130	140	150	170
Fresh wt. (g)	seed corm	65.0	58.5		10.7								
	new corm				6.7	17.5	50	81	152	234	365	408	382
Dry material/ fresh wt. (g)	seed corm	20.0	18.2		8.6								
	new corm				11.2	18.8	20.0	20.2	20.6	20.0	14.4	13.5	16.5
Dry material (g)	seed corm	13.0	10.6		0.9								
	new corm				0.7	3.3	9.2	16.5	31.7	46.8	52.6	55.2	57.3
Daily increased dry wt. (g)	seed corm	–0.1		–0.3									
	new corm					0.3	0.4	0.7	0.8	0.8	0.6	0.3	0.1
KGM (%)	seed corm	51.5	41.9		7.2								
	new corm				24.3	36.7	50.9	51.8	56.0	55.3	54.3	54.5	53.2
Starch (%)	seed corm	10.5	7.5		1.0								
	new corm				1.3	8.3	10.9	10.7	10.6	10.8	10.5	10.9	10.7
Soluble sugar (%)	seed corm	4.8	5.6	8.9	6.8								
	new corm				6.9	8.7	6.4	6.2	5.8	5.4	4.9	5.2	4.8
Protein (%)	seed corm	8.9	11.8	14.0	6.5								
	new corm				13.4	7.1	6.0	5.6	5.0	5.8	7.0	7.5	8.0

Table 9. The Decrease Percentage of Corm Component after 4 Months Storage

Storage temp.	Fresh weight	Dry weight	KGM content	Starch content	Solublesugar content	Rough protein content *
5℃	20.0	8.8	3.7	18.9	0.07	9.4
10℃	17.7	5.5	1.8	13.9	0.04	5.7
20℃	22.2	12.7	3.9	38.9	0.09	16.5

* to show the increased percentage

(4) β – mannans

Shi *et al*. (1988), studied the β – mannans activity and property in the corms of *A. konjac*. Throughout the dormancy stage β – mannans activities were present and relatively unchanging in the corms, but in the ger-

minating corms β – mannans showed a rapid increase in activity. In the new corm forming stage, at first the activities were relatively high and then decreased somewhat in the new corm.

The purified enzyme showed optimal activity at pH5.3, optimum temperature of 40℃, it is considerably stable at pH4.0 – 8.6. As the K_{mox} and max values for konjac mannan, 4.2×10^{-2}(%) and 9.1×10^{-2} (0.0560nm) were obtained, respectively.

(5) Research on the Properties of Glucomannan

The refined powder of konjac is a kind of rough glucomannan(GM). The result of research (Jia *et al*., 1988), showed that glucomannan were composed of D – glucose and D – mannose, the molar ratio of G_1 and M in different species were not the same. It is 1:1.69 in *A*. *albus* , and was 1:1.6 *A*. *konjac* . Their molecular weights were estimated to be 8.09×10^5 and 7.37×10^5 respectively. The D – glucose and D – mannose residues of both GM were joined together by β – 1, 4 – linkage. It seemed that some sugar residues had an o – acetyl group.

The chemical structure and the rheological properties of GM from *A*. *albus* and *A*. *konjac* were studied by Xu *et al*. (1991). The X – ray diffraction data showed that grains of these KGM were amorphous, so the swelling capacity of the GM is very strong, and the aqueous solution of KGM is a typical pseudoplastic fluid.

Yu (1990), Liu (1992), and Hu *et al*. (1990), carried out studies on the graft copolymerization of acrylonitrile (AN), or butyl acrylate onto konjac glucomannan, and the esterification reaction of phosphate onto KGM. The results of chemical modification of KGM show that the KGM polyacrylonitrile graft copolymers have higher viscosity, 2 – 4 times that of KGM. The relative stability of the sol. Of graft copolymer increased from 22% of KGM to 84%, and the membrane of the graft copolymer is more uniform and more tight that that of KGM.

5. Research on Biotechnology of Konjac Plants

(1) Microproduction

The reproduction rate of konjac is very low, therefore it is difficult to quickly extend new varieties. Huang (1992) and Zhang (1993) researched the microproduction *in vitro* of konjac. They established the microproduction system and furthered research on the mechanism of cell differentiation and the formation of organs.

In addition to the common way of shoot formation, another four ways of organogeny were found: which are root formation before shoot formation, small corm formation, embroil formation and dormant bud of explain developing into plantlets directly.

Embroil formation of konjac was special *in vitro* . The embroils occurred in the surface of calli, cutting for bud – scale or in the internal part of calli. The former developed from cells of occurred. But the latter originated from a single cell proembryo cell, its division formed bicells of the original embryo, then it divided disorderly and developed into a cormel which showed obvious cormel structure. It is the same as the developing process of sexual embryo of konjac.

(2) Artificial seed

In non – sterile soil the germination rate of artificial seed encapsulated with several antibiotics was 12.5%, two times that of artificial seed without antibiotics.

(3) The isolation and culture of protoplasts of konjac

Protoplasts were isolated enzymatically from young leaves, bud scales and petioles of *A*. *albus* and *A*. *konjac* . High rate of division of protoplast was gained in modified BSB, D2a or MS liquid media with or with-

out the addition of NAA, 2,4 – D, BA, KT, or GA_3. The first divisions of protoplasts were normal equational divisions, but the following divisions were not in sequence, no embroil were observed. Browning is a serious problem in the culture of protoplast of konjac, supplementation with 0.1% PVP and 0.1% Vc in the media couldn't prevent it.

RESEARCHES ON AGRONOMY OF KONJAC

1. Planting Demarcation of Konjac

Yang *et al*. (1990) obtained a series of temperature indices by temperature experiments on *A. konjac* in the auto – controlled artificial growth chamber according to the schedule of 15/10, 20/15, 25/20, 30/20, 40/30 and 45/35. The ecological and geographical conditions affecting the growth and development of konjac plants were investigated. It was found that the heated and water conditions are the key factor and the geographical location, altitude, venation are the indirect key factors. Sixteen climate indices were selected as the standard index system. Two hundred sets of meteorological materials from 12 provinces of China were joined in Fuzzy clustering analysis. The result of Fuzzy clustering considerably conformed to the distribution of konjac germplasm resources in China.

According to Fuzzy clustering and suitability for planting konjac, the syntheticly country was demarcated into 4 main regions, consisting of 6 subregions of China.

2. Efficient Cultivation System

The peasants need both food and income. To realize this purpose, Liu *et al*. (Southwest Agricultural University) have researched and created a new system of intercropping konjac with high – straw grain crops, such as maize and sorghum, growing in the upper space where they can take in sufficient sunlight without a reduction in output. Between the rows, konjac is inter – planted between 2 or 3 rows growing in the lower space where they can be in a position of proper sun and shade. This reduces physiological and pathological diseases, and gives higher yields and income.

In 1980s Liu *et al*. extended this system in Wanyuan County, Sichuan Province in Daba Mountains area where the peasants already changed from the old custom of random cultivation into linked vast cultivation. In 1990 the cultivation area had developed to 400 hectares. At the same time Liu *et al*. helped the peasants to dry fresh konjac corms into pieces, to process konjac pieces into fine powder, and to set up snow white konjac processing plant. Thus, konjac cultivation and processing has become a new industry which enabled the peasants to surpass the standard of having just enough to eat and wear, and by which the peasants have been free from poverty and are becoming richer. From Wanyuan County the experience was extended to Sichuan Province and to the whole nation, and the cultivation areas have been exceeded 7,000 hectares and 30, 000 hectares respectively.

3. The Breeding of Konjac

The breeding of konjac has three objectives; (1) raising the yield per unit area, (2) increasing KGM (konjac glucomannan) content in konjac corms, and (3) improving the anti – disease capacity. The breeding of konjac has two methods, one is selection breeding, the other is cross – breeding. The yield and corm composition are quite different even for *A. konjac* in the different parts of China. This shows that there is selection potential for various areas. Liu *et al*. have selected out Wanyuan *A. konjac* and has passed breeding examination. Because konjac has for long time been reproduced asexual, mutation and variations can be easily passed on. There possibly exists variation even for plants reproduced from the same mother corm by cutting, so there exists the possibility for field individual selection.

The characters of high yield, high KGM content and high anti – disease have never been found in the

plants of same species but have often been found in different species, therefore, it is necessary to adopt the way of cross - breeding. Interspecific crossing has an especially high potential. But interspecific cross still has some problems in techniques on which Liu *et al*. has done much research work and has been successful in solving the following problems.

(1) The long period of breeding

Konjac needs nearly 4 years to blossom. In order to shift flowering to an earlier time, Sun and Zhang *et al*. (1988), applied the proper concentration of GA_3. The pistil produced by this method can develop normally and be fertilized and bear seeds, but the problem of male sterility still remains unsolved.

(2) The different flowering time of different species

The flowering time of different species is sometime separated by too many days. According to Zhang's experiment, low - temperature storage of early - florescence species can postpone its time of flowering in order to meet the time of late - flowering species.

(3) Protogynous flower and isolation of pistil and stamen

The female flowers mature 2 days earlier than male flowers of the same plant and lose fertility rapidly. The pollen has to be collected from the blossomed plant and stored for further use in interspecific hybridization. The male flowers on the upper part of a inflorescence cannot be emasculated after artificial pollination. The best method to solve this problem is to daub the male inflorescence with Vaseline and seal it so as to prevent it from releasing pollen, then bagging it for several days. Relative humidity should be maintained at 80% in the periods of pollination and seed - bearing.

(4) The propagation and spread of fine varieties

The propagation coefficient of konjac is so low that breeding a fine variety by normal method needs at least 10 years and spreading a variety at least 20 years. Zhang *et al*. (1993) has already set up a system of tissue culture micropropagation which anticipate will reduce in half, the time needed.

STUDIES ON THE MEDICAL EFFECT OF KONJAC

Early in 1986 the Western China Medical University (in Chengdu City) and the Third Chinese Military Medical University (in Chongqing City) began to study the medical and hygiene effect of konjac to human body.

The main composition of konjac is glucomannan KGM which is a kind of fluid - semicellulose and can be used to control and alleviate corpulence and to prevent constipation. Liang who works in the Western China Medical University studied the effect of KGM on the glucolipid metabolism of diabetes patients by using konjac KGM to treat noninsulin - dependent diabetics. After one month of treatment of three times a day (2.4 grams altogether), the patients' blood sugar content decreased 18% and 10% in empty stomach and after - eating respectively. Total cholesterol decreased 19%, and glyceryltriester decreased 30%. High intensity fat protein increased 13%. However, konjac KGM can be used as a subsidiary drug to treat diabetes and high blood fat disease.

Peng *et al*. (1994) conducted research on the effect of konjac fine powder to inorganic nutrition, and to the removal and blockade action of harmful inorganic ions. It has no effect on the apparent digestive rates of calcium, iron and zinc on the human body. Eating 9 grams konjac fine powder a day. There is no notable difference between the weight of femur, content of calcium and phosphorus, the measure parameter of bone shape, with the control group after eating for 18 months. In the simulated experiment of gastrointestinal envi-

ronment, konjac fine powder was found to combine with Pb2 +, Cd2 +, Zn2 +, Ca2 +, and Fe2 +. Chemical examination of excrement and urine showed that fine powder can exclude and blockade lead absorption of human body, but cannot remove absorbed lead. However, fine powder can release the cadmium poisoning reaction of rats.

KONJAC USAGE BY PROCESSING IN CHINA

1. Konjac Pieces Drying

The konjac cultivated randomly by Chinese farmers is processed as konjac curd from fresh corms, or is dried into konjac pieces to sell, whose quality is not very good. Since the 1980s there became a problem of technology and equipment for konjac drying because of the expanding of cultivation area and the increasing of yield. Liu *et al*. found two ways to solve this problem. One is to improve the farmers' drying solution methods in order to prevent the contamination of pieces by the smoke form burning coal, and to change the drying method from direct drying into indirect drying method. The other is the creation of 6YMHW – 200 and Z – 400 type drying equipment which uses hot wind stoves to blow hot air for dehydration, controls temperature and humidity automatically in order to prevent browning and improves the processing quality. The Chinese konjac dried pieces processed by this way have entered the international market.

2. Fine Powder Processing

In 1986 Southwest Agricultural University and the 317 Research Institute of Spaceflight Ministry were successfully made a MJJO – 01 type konjac fine powder machine using dry production technology. Konjac fine powder made first by Chinese equipment came into the international market. Up to now 80% of Chinese konjac fine powder processing plant still use this machine for its low cost and high effect.

Chen *et al*. (1986) researched alcohol wet production technology successfully. Li *et al*. (1992) finished the technological and equipment design, and it is used in real production since that time.

3. Usage in the Processing of Food and Beverages

(1) Food

a. Konjac curd can be braised with duck, chicken meat because it has a high force of absorbing flavor, e.g., "konjac braised with duck" has become a traditional Chinese dish which can also be processed as canned food.

b. Taking advantage of the accumulation of snow and low temperature in Emei Mountain, konjac curd can be further processed into honeycomb – like snow konjac which has a higher force of absorption flavor and can be made into various dishes. Sun *et al*. (1986) were successful in making artificial snow white konjac in a freezing room and set up two snow konjac processing plant in Wanyuan County.

c. Konjac curd can be further properly dehydrated and puffed, adjusted in flavor and color and made into various foods such as astringent and hot dried beef.

(2) Since 1980's, various features of hydrophilia, thickening, stability, emulsifiability, suspending, coagulability and film – forming of the glucomannan in konjac had been made use of in developing new kinds of food and is used as an additive in beverage procession in China.

a. Strengthen the tenacity of noodles and bean starch vermicelli, reducing rate of breakage, without sticking together and troubled water, making them smooth and with long storage time.

b. Used as an additive for biscuits, bread, cakes and buns etc. wheaten food to make their outward appearance glossy, their texture spongy, and to prevent them from aging, to be moisture proof, and to prolong storage time.

c. Used as stabilizer for ice – cream to make the materials stable and even without forming ice crystals and makes the taste fine and smooth. The concentration is 0.1% – 0.3%, lower than other stabilizers, but the effect is better.

d. Increasing the intensity of anti – pulling of glutinous rice paper.

e. Used as a stabilizer for the head on a glass of beer to make the foam exquisite and even and maintained for a longer time.

f. Used as a suspending agent for fruit juice with flesh, vegetable and tremella.

g. Used as clarifying agent for alcoholic drinks and fruit juices.

h. Food preservative Immersing eggs, fish, meat, fruits etc. in 0.5% – 1% konjac fine powder solution because the existence of ions can form a layer of plastic – like film which has a high permeability coefficient for CO_2 and low for O_2 and which can reduce transpiration, inhibit the propagation of aerobic microorganisms, and prolong the fresh – state period of food.

4. Usage in Other Industries

(1) Cosmetics

Glucomannan in konjac has a wonderful hydrophilia and film – forming feature, which is helpful to protect skin and hair. It prevents skin and hair from losing water and can block direct sunlight. A series of shampoo, hair oil, skin – lubricating cream and sunscreen have been developed since 1980's.

(2) Drilling Fluid

Zeng (1989) researched the application of KGM gum as non – clay drilling fluid. It is especially suitable for diamond drilling, drilling for all kinds of downhole motors, hydrogeological wells and water well drilling and shallow oil well drilling. It possesses many excellent properties than other drilling fluids. KGM fluid has been used for drilling in North – China oil wells and Sichuan oil wells, but a new problem is that the KGM gum increased the production cost.

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